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PRELIMINARY REPORT OF APHIS INFESTATION OF PONDEROSA PINE ON THE PRESCOTT NATIONAL FOREST, ARIZONA DURING THE 1935 SEASON

by
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Berkeley, California
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PRELIMINARY REPORT OF APHIS INFESTATION OF PONDEROSA PINE ON THE PRESCOTT NATIONAL FOREST, ARIZONA, DURING THE 1935 SEASON

INTRODUCTION

During the late spring and summer of 1934 an excessive abundance of a stem feeding aphis on ponderosa pine occurred in the Prescott National Forest, Arizona. Some attention was attracted to this insect because of its abundance and also because of the prevalence of a twig killing blite in the forest. There was the existant possibility of some connection between the blite and the insect.

According to available information, the greatest abundance of the aphid was noted in the last part of May and the first part of June in 1934. By the end of the latter month the aphis had practically disappeared in the immediate vicinity of the city of Prescott (elevation 5200 feet), but were still to be found in considerable numbers on Mingus Mt. at an elevation of 7500 feet. That a heavy parasitism occurred in several localities near Prescott was evidenced by the dead bodies which clung to the branches even during the 1935 season.

Observers reported that during 1934 the aphids were everywhere abundant on ponderosa pine, covering the branches from small twigs to the main trunks, and secreting "honey dew" in sufficient quantities to coat the ground with an encrustation.

Because of this condition and the dearth of information on pine infesting aphis, a field station, under the direction of the Berkeley Office, was set up in the Prescott Forest on May 9, 1935. Living quarters and a field laboratory were located at Civilian Conservation Corps Camp F-62-A, 13 miles outside the city of Prescott, and in an area of the forest where a heavy abundance of the stem aphid had been noted in 1934.

The purpose of this investigation was to determine what species of aphids were present, to secure as much information as possible regarding their life cycle and feeding activity, and to consider the possibility of some relation between twig blite and aphis activity. Unfortunately, the 1935 season has been a poor one for aphids on the Prescott. This fact, coupled with the necessity of curtailing the field program owing to financial exigencies has made it impossible to secure all the desired information. Specimens of species found in the forest were collected and have been identified. Attempt was made to rear these forms under caged conditions and field observations were carried on through the season.

Aphid Species Found on the Prescott National Forest in 1935

The aphids collected during the field season have been identified by Professor E. O. Essig of the Department of Entomology, University of California. In all, four species were collected, one of which was the form so prevalent during the 1934 season.

The following species were identified:

Schizolachnus pini-radiatae, a needle feeding form; black, with striped abdomen; long legged.

Cinara sp., a second needle feeding form; greyish; heavily pubescent.

Chermes sp., a woolly aphid, feeding on new growth.

Cinara schwarzii, the stem feeding aphis. This species was first described by Wilson* as Lachniella schwarzii from material at Laporte, Colorado in 1898, and also Prescott, Arizona in 1901. Gillette and Palmer** list the species as Cinara schwarzii (Wilson). The adult form is described by the latter authors as: "Cinnamon brown, mottled with black to entirely black; with scattered patches and streaks of pulverulence; legs mostly blackish; cornicles black". In Colorado the species is common wherever the host (Pinus ponderosa var. scopulorum) occurs on the eastern slopes of the Rocky Mountains. In that locality the appearance of the various forms was noted as follows: fundatrices, April 8 to May 18; apterous summer viviparae, June 18 to November 8; elate viviparae, June 18 to October 7; sexuales, October 9 to 27.

Resume of 1935 Conditions

At the time when the field station was established (May 9), one species of needle feeding aphis was present in the vicinity of camp. This was Schizolachnus pini-radiatae, which was found to be generally distributed in the forest but nowhere exceedingly abundant. The numbers of this species decreased as the season advanced, and from the middle of June onward, only a few scattered colonies were located from time to time.

A second needle feeding form, Cinara sp., was also found to be generally distributed but more discontinuously from the middle of May into June when it likewise decreased in numbers. After the middle of July an increase in this species was noted but the distribution was even more sporadic than earlier in the season. At no time could

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* Wilson, H. A. 1919: "Some new lachnids of the genus Lachniella." Can. Ent. 51: 41.

**Gillette, C. P. and M. A. Palmer. 1931: "The Aphididae of Colorado." Ann. Ent. Soc. Amer. 24: 868-9

this form have been considered abundant.

The presence of a woolly aphid, Chermes sp., in considerable abundance on the new growth tips was noted at the time when the field station was established. In a few localities this species was heavily infesting young trees, but after the middle of June the numbers lessened materially and during July only a few scattered colonies were found.

Cinara schwarzii, the aphid which was the primary object of the investigation was first found on June 17 in the immediate vicinity of the camp. Subsequent scouting located this species in one other area near Prescott, and on Mingus Mt. on June 25. The greatest abundance of this species was found between the first and tenth of July. During the latter part of June there was a sudden decrease in the numbers of this insect and at the time of closing the field station on August 1, only a few infested trees could be located.

Rearing Experiments and Field Observations

For the purpose of rearing under observation, several cages were built of muslin over a chicken wire frame. These cages were three feet in length and approximately one and one-half feet in diameter. They were placed over the branch of a tree and secured at either end. A certain degree of observation without opening the cage was made possible by inserting a small screen window in the two sides of the cages. Since, however, this method did not allow close observation without opening the cage, a larger cage was constructed enclosing the whole tree. The dimensions of this structure were eight feet on a side, and a tree slightly less than eight feet in height was enclosed. With approximately 200 tips on the tree, ample material was available for experimental work.

It was found that the temperature inside this cage rose above that of the outside air. To remedy this situation a two and one-half foot square window of mosquito bar was inserted in the muslin wall of each side of the cage. In addition a canvas awning was stretched over the top. Under these conditions the temperature inside the cage closely approximated that of the outside air.

Needle Feeding and Wooly Aphis

All attempts to rear both forms of needle feeding aphis and the woolly aphid were unsuccessful. These forms were transferred individually with a brush to tips on the caged tree, and also infested tips collected in the field were lightly tied to tips in the caged tree. Where the transfer was artificially accomplished the aphids died in a few days. Where the transfer was left to the insects themselves, they died on the tips collected in the field without moving over to the fresh material which was available. According to information secured from Professor Essig, the needle feeding aphis are characteristically unable to adapt themselves to a new medium once they have commenced feeding.

Stem Aphis

When the stem aphis first appeared, introduction to caged tips was accomplished by transferring individuals with a brush. Later, more successful colonies were established by bringing the infested tips in from the field and tying them to tips in the caged tree. The aphids then made the transfer on their own volition, as the tip on which they had been feeding dried out.

All of the infestations found in the field at first were on the current year's growth. Accordingly, cage introductions were made to the 1935 growth. The adults, summer viviparae, appeared to most readily make the transfer, whereas the younger individuals for the most part died on the stem on which they had been feeding in the field. Approximately 50% of the attempted colonizations were successful. In all, the current year's growth of 119 tips was infested during the season from June 19 to August 1. During the latter part of this period when the aphids fed on the older growth, some new infestations of the 1935 growth occurred due to migrations of individuals from one tip to another.

During the last part of June the greatest mortality among introduced colonies occurred. Primarily this was due to the viviparae dying without producing young. During the first week of July, the production of young materially increased and reached a peak about the 10th of July. The greatest average production of young in any colony over a 48-hour period was 6 per female. The total production of young was of course much greater, occuring during the last half of July. It was during this period that migration backwards onto the older stems occurred.

On July 29 several colonies were started on older twigs from material collected from corresponding locations on trees in the Coconino National Forest near Flagstaff. With one exeeption these colonies were successfully established. During the first part of July it became apparent that the aphids were feeding more toward the base of the current year's growth, and many instances were noted where the aphids were clustered about the node between the 1934 and 1935 growth. About the middle of July further migration took place and approximately 25 colonies were established on the 1932, 1933, and 1934 growth. Several branches were infested from the growth of the current year back onto the 1932 growth. Conditions on the older growth were apparently more favorable to the aphids which greatly increased in numbersa and literally covered the infested branches. The feeding colonies consisted practically of a contiguous mass of aphids, but within this mass it was noted that the individuals were grouped into somewhat irregular rosette formations. Figure 1 is a photograph of several of these colonies. The branches were cut from their normal position and arranged for the purpose of photographing. In the course of this procedure many of the aphids either fell or were brushed off the branches. Hence, a true picture would show a density all along the branches comparable to the greatest abundance shown in the photograph.

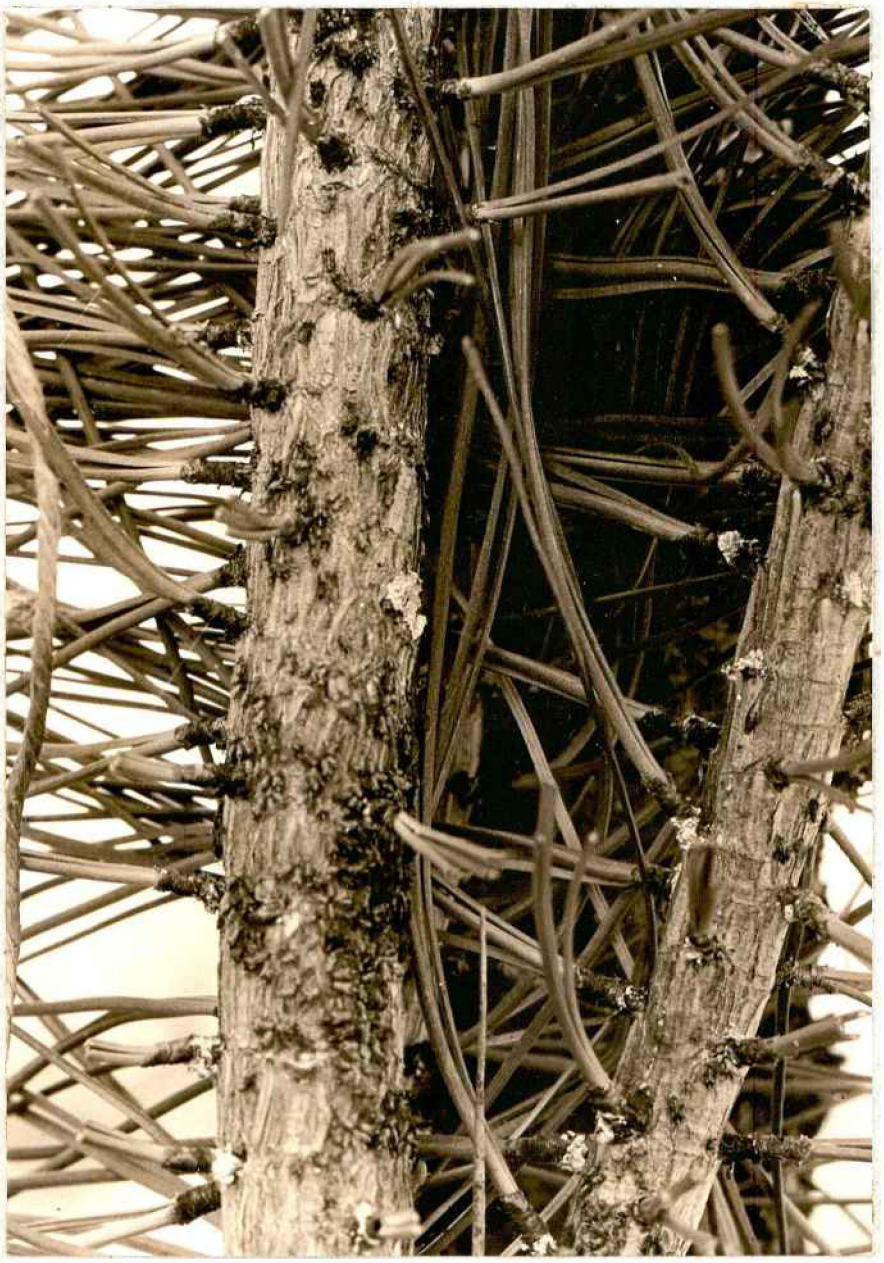

Figure 1. Stem Aphis, Cinara schwarzii, on Pinus ponderosa, Prescott National Forest, August 1, 1935. Photo by J. E. Patterson No. 9362

No distinct preference for the twigs of any one year was noted, and the manner in which migrations took place did not indicate that the individuals were confined to a particular year's growth. In the majority of cases where a colony was spread along a branch the greatest concentration occurred on the growth of the 1933 season.

During the last week of July the production of winged forms, alate viviparae, materially increased. In some cases these individuals remained feeding in the colonies, but were commonly found resting on the needle tips or on the walls of the cage. When several groups of these winged forms were confined on a section of branch they produced young and started new colonies.

Field Observations on Needle Forms and Woolly Aphis

Both forms of needle feeding aphis and the woolly aphis were found in a number of localities in the forest. All three forms were confined to young trees, 10 feet or less in height. No continuous distribution of any of these forms occurred over an extensive area, nor were they present for a protracted period at any one place in the forest. The woolly aphis was abundant on groups of reproduction during May, while the new growth was budding forth. Concurrently with the elongation of the terminal shoots this aphid decreased in abundance. No evidence of any serious effect of the attack was observed. A number of instances of dead tips occurred where both an aphid and a tip moth infestation were present. In all probability this killing was due to the activity of the latter insect.

Of the two needle forms, Schizolachnus was the more common, but Cinara was more abundant on individual trees, particularly during July. A count of five trees infested with the latter form showed an average of 49 needles colonies per tree, with extremes of 20 and 100, and an average of 8.8 individuals per colony, with extremes of 1 and 35. In the case of Schizolachnus no more than 20 colonies were found on any one tree, although the size of the colonies was approximately the same as those of Cinara.

Field Observations on Stem Aphis

As previously stated, field conditions during the 1935 season were unfavorable for a heavy stem aphis infestation. The factors making for a decrease in abundance of this species as compared with the 1934 season are unknown. Certainly climatic influences were important and possible parasites and predators played a role. Of the three localities where infestations were found in 1935, only in the Lynx Creek region was the stem aphid present until the first of August. At that time the colonies in this district were very few in number.

On July 10 a survey was made of young trees in the vicinity of the field laboratory. Of 86 trees examined, 27, or 31.4% were infested, with an average of 3.9 infested tips per tree. During the period July 17-19 a similar survey was made along Lynx Creek southward from the camp, examining young trees on either side of the road. This survey strip was approximately four miles in length. Of 884 trees ex-

amined, 49, or 5.5% were infested, with an average of 2.5 infested tips per tree. In size these trees ranged from 2 to 12 feet. Infested trees in the Peterson area and on Mingus Mt. were of the same size. With one exception, no colonies were found on the older and larger trees. This was in distinct contrast to 1934 conditions. In that year trees of all sizes were heavily infested and from the larger trees a steady drip of "honey dew" took place. Nowhere was this condition observed in 1935. The single exception alluded to above was with reference to two small colonies located in the Hassayampa Creek Territory. In both cases the colonies were found on lower branches which were in the process of being naturally pruned.

During the third week in June a number of infested trees were tagged in the infested areas. Frequent visits were made to these trees for the purpose of checking the development of colonies in the field. Through the first week in June there was a gradual increase in the number of infested tips on the trees as well as the size of the colonies themselves. Nowhere were more than a dozen infested tips found on a tree. During the second week of July the numbers sharply decreased and in the third week the aphids disappeared entirely with few exceptions. This sporadic appearance and disappearance was characteristic all during the season. In several cases infested trees were tagged and within three days the infestations had entirely disappeared.

Some mention should be made of the ants which were invariably associated with aphid colonies on the stems. Two ant species were present, the smaller of which was more generally distributed. The ants were usually active about the colony and many individuals were observed stroking aphis with their antennae, apparently to encourage the exudation of "honey dew." Whenever the aphis colony was disturbed by moving the twig or in any other manner, the ants immedia tely became very excited. The larger species would attempt to attack a pencil or object which was moved near the aphis.

Cinara edulis

On the 9th of July, 1935, specimens of a stem aphis infesting the branches of Pinus edulis were collected in Little Walnut Canyon on the Gila National Forest, New Mexico, by Messrs. Frink and Sandberg of the Prescott Office of Forest Pathology. This form was determined as Cinara edulis, closely related to C. schwarzii, and only differentiated from the latter because of the accepted belief of specificity of host for members of the genus Cinara. A colony from this material was placed on the cage P. ponderosa tree but was not successful. The reason for this may have been due as much to the poor condition in which the aphids arrived as to their inability to make the change of hosts.

Biotic and Climatic Influences

Because of the brief seasonal appearance of Cinara schwarzii, it is difficult to determine what facotrs, climatic and biotic, may influence fluctuations in aphid populations. It is evident that some factor or group of factors, possibly a combination of climatic and biotic, brought about a sharp reduction of the epidemic which existed in 1934. The possibility is strong that in certain areas of the forest parasites played an important role, as is evidenced by the remains of the parasitized bodies

which were found on stems heavily infested during the 1934 season. In one of these areas the remains are exceedingly abundant, and certainly for that particular region parasites were one of the chief factors contributing to the decline in population. A slight degree of parasitism was noted in 1935. In all three infested areas occasional parasitized bodies were found, but nowhere in sufficient abundance to have accounted for the sudden disappearance of the colonies. An Ichneumon and a Chalcid species were reared from parasitized material which was collected in the field.

Of possible predators, lady-bird beetles (Coccinellidae) were exceedingly abundant. Numberous reports and observations were made of cases where the beetles were clustered on branches of the pine trees or on rocks literally by the thousands. Whereas no actual predation was seen, these beetles are known to feed on aphis. In 1934 lady-birds were present in many areas where heavy aphid infestations existed.

In the latter part of May a considerable abundance of a species of Syrphid fly (Syrphidae) was noted. Although this was before the stem aphis appeared, a large group of Syrphids are predatory on aphis and scale insects, and the species present on the Prescott would probably feed on the aphis where a heavy infestation existed. A third possible predator commonly found on ponderosa pine was a serpent fly of the genus Rhaphidia, probably Rhaphidia arizonica.

Sudden fluctuation in aphid populations are notably characteristic of this groupbof insects. Climatic influences are important influences determining the rise and fall of epidemics. On the basis of a single season's observations it is impossible to determine any specific climatic effects. The rainfall factor may possibly limit aphid abundance. The decline of population on the Prescott immediately followed the first hard showers of the rainy season in July. In 1934, however, the peak of abundance occurred before the end of June and before the advent of the rainy season.

The following weather records are included for reference purposes. No interpretation in light of these records is made regarding the fluctuation in aphid abundance. The data were recorded at the U. S. Veterans' Hospital in Prescott, Arizona.

MONTHLY MEAN MAXIMUM-MINIMUM TEMPERATURES IN ^{o}F

	May		June		July	
	Max.	Min.	Max.	Min.	Max.	Min.
1934:	84.7	44.2	84.9	43.5	94.3	58.0
1935:	72.8	38.0	97.7	47.7	92.6	63.2

MONTHLY PRECIPITATION IN INCHES

	May	June	July
1934:	1.65	0.15	0.63
1935:	.13	0.00	2.37

The stem aphid overwinters in the egg stage on the needle of the host tree. The possible effect of winter temperatures and mositure conditions are of course unknown.

Aphis and Twig Blite

The possibility that there can be any connection between the stem aphis and the twig blite which is infecting a large are in the Prescott Forest seems remote. Whereas some justification for such a theory may have existed in 1934 when the aphis was abundant, it is difficult to support such an argument in light of the scarcity of the aphis during the current year. The area infected with blite has increased in the past year. Blited areas exist where there is no evidence of a heavy aphis infestation previously, and it is definitely shown by recent inoculation studies that a fungus present in the blited twigs is the cause of the injury.

As to what effect a heavy aphis feeding has on the host tree, further study will be necessary to determine. The theory that an aphis attack in one year might cause an injury similar in appearance to that caused by the blite organism to appear even the following year is not substantiated. Branches which were heavily infested in 1934 are perfectly normal in appearance in 1935. This is true both in blited and unblited areas.

Observation of aphis conditions during the remainder of the current season and location of the eggs in the fall would be of value. General observations are being made by an enrollee at the camp where the field station was located. This will furnish a check on any activity of the species which might occur although it is believed that the season for this year is now past.

HISTOLOGICAL STUDIES

In order to substantiate, if possible, the conclusions drawn from field studies, and to determine if any changes in wood structure result from an aphis infestation, a histological study has been undertaken. In addition to sectioning twigs which had been attacked by aphis, normal twigs and also blite infested material was sectioned. By this means any abnormal cellular proliferation, occlusion or other apertrophy could be compared with normal tissue and with the effects of fungous invasion of ponderosa pine stems.

Material and Technique

All material was preserved at the time of collection in an acetic acid-alcohol-formalin solution. Most of the specimens were transferred to glycerin following fixation since the latter is a better medium for storing tissues.

For purposes of references the material examined is tabulated below, together with method of cutting, staining and certain annotations:

No.	Condition of Infestation	Microtome	Stain	Notes
P-1	Heavily infested in '34 a. 1927 origin b. 1928 origin c. 1929 origin d. 1930 origin e. 1931 origin	Sliding	Safr.-Del. Haem.	Material cut from both glycerin and 40% alcohol. Twigs from lower branches of 50' tree.
P-2	Heavily infested in '34 a. 1925 origin b. 1926 origin	Sliding	Safr.-Del.Haem.	As above
P-3	Uninfested '35 origin	Sliding	Safr.-Del.Haem.	From 6' tree in cluster of reproduction,
P-4	Uninfested	Sliding	Safr.-Del.Haem.	From lower branches of 40' tree.
P-5	Infested '35	Rotary	Safr.-Del.Haem.	From caged tree mostly '34 and '35 twigs.
P-6	Blited a. 1st internode b. 2nd internode c. 3rd internode d. 4th internode	Sliding	Safr.-Del.Haem.	From blite area
P-7	Infested '35	Rotary	Safr.-Del.Haem.	1935 origin; from 6' tree

As noted in the preceding table all material excepting lots P-5 and P-7 were cut on the sliding microtome. For this work no embedding was necessary, and fair sections for the most part were obtained at 24 microns. The specimens in lots P-5 and P-7 were transferred to tertiary butyl alcohol, a procedure which eliminates the use of an ethyl series with its consequent hardening effect, then through a mixture with soft paraffin to soft paraffin and was embedded in hard paraffin. From the paraffin blocks thus obtained, ribbons were cut at 5 microns. The reason for embedding these specimens was that aphids were still clinging to the twigs, and it was hoped to secure sections with the aphids in situ.

In all cases the sections were stained in an alcoholic safranin solution and counterstained in Delafield's Haematoxylin. Stylet tracts will take up the safranin stain more deeply than surrounding tissues, in most cases. Davidson reports, however, that frequently these tracts will not

stain at all. A few sections were treated in acidulated alcohol (2% HCl in 95% alcohol) for 24 hours and stained with aqueous methylene blue, but less satisfactory results were obtained by this method. The nature of demonstrating the sytlet sheathes by this method is to suggest the presence of pectic substances.

Sections cut on the sliding microtome were mounted in glycerin jellow following staining. Paraffin ribbon sections were placed on slides, stained, and mounted in Canada balsam.

All photographs were made with the assistance of J. E. Patterson.

Literature

The literature dealing with the effect of sucking insects on forest trees is scant indeed. Although an exhaustive search has not been made, only a few papers related to this problem have been uncovered. These are briefly reviewed in the following paragraphs.

The work of Davidson (1923*, 1925**) is of considerable value, although the studies reported do not include any work on aphids affecting conifers. Some of the generalities, however, are applicable to the present discussion. The aphids obtain their food by penetrating the host tissues by means of a flexible, chitinous piercing organ composed of the maxillary stylets and mandibles. The former are closely opposed, resulting in the formation of two canals. The dorsally situated one is the suction canal through which the plant juices are conducted into the pharynx. Ventrally lies the ejector canal down which the saliva is pumped into the host. Penetration of the piercing organ is brought about by retraction of the labium coincidentally with a prolongation of the "forehead" with the labium gripping the distal end of the piercing organ in pincer-like fashion. Davidson believes that the saliva has a two-fold function; that of dissolving a passage for the piercing organ, and also the ability to bring about the conversion of starch to sugars. A well marked stylet sheath is formed about the piercing organ as it proceeds into the host tissue. This sheath is probably composed of callose together with insoluble calcium pectate and a deposition of tannin. Reacting on the cell contents the saliva causes plasmolysis and consequent disorganization. In the case of Macrosyphum on rose Davison found that the salivary reaction brought about a cellulose thickening of the affected cells. In extensive studies with Aphis rumicis on various hosts it was shown that but little spreading of the irritating effect of the salivary fluid to surrounding cells occurred. Davidson believes that the action of this fluid is more important than the piercing.

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* Davidson, J. 1923. "The penetration of plant tissues and the source of the food supply of aphids." Ann. Appl. Biol. 10: 35-53.

** Davidson, J. 1925. "Biological Studies of Aphis rumicis Linn. Factors affecting the infestation of Vicea faba with Aphis rumicis. Ann. Appl. Biol. 12: 472-507.

Busgen* (1891) believed that the sheath was composed wholly as a product of salivary secretion rather than in combination with the plant juices, and that this structure acted as a support for the stylets. The demonstration of the presence of pectic substances in the sheath, as well as other tests, indicate that the plant juices are involved in the sheath formation. The fragility of the sheath in comparison with the stylets makes the support thesis somewhat untenable.

It is evident from the discussions of Davidson and others that pathological features of the host may be exhibited as the result of an aphis infestation, particularly as caused by the salivary action. To quote from Davidson: "However, it must not be overlooked that once the physiological relationship existing inside the cells are disturbed owing to the influence of the saliva, the complex chemical changes which may result cannot at the present stage of our knowledge be clearly understood/ Further research is necessary on the action of saliva of different species of aphids on different plant hosts and the changes which occur in plant tissues infected with salivary secretion. Further, the physiology of digestion in aphis requires to be investigated." This statement clearly expresses the lack of knowledge concerning such a problem as the present one.

In an article on a comparative study of the feeding methods of certain Hemiptera, Smith** (1926) states that in aphids the penetration of the plant tissues is generally intercellular in nature with the creation of a well defined stylet tract. Less actual mechanical damage is caused by aphis feeding punctures than by certain other Hemiptera which have a barbed rostrum. Smith also concurred that in the case of aphids there was little or no damage caused by the salivary secretion whereas the salivary secretion of Capsids, for example, has a violently toxic effect on the plant tissues.

Considerable controversy exists in regard to the manner in which the plant sap is obtained. Busgen classified the course of the stylets in the plant tissues as inter or intra-cellular. His figures of Lachnus sp. occuring on Picea alba show the course of the stylets as intracellular, in part at least. Zweigelt***(1915) considered inter-cellular passage of the stylets to be associated with extensive inter-cellular suction as the principle method by which the cell sap is obtained. Davidson (1923) on the other hand, believes that intra-cellular suction is most important, sponsoring the suggestion that the cell membrane would exert

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* Busgen, M. 1891: "Der Honigtau." Jena Zeitschr.f.Naturwiss XXV:340-428

**Smith, K. M. 1926: "A comparative study of the feeding methods of certain hemiptera and of the resulting effects upon the plant tissue with special reference to the potato plant." Ann. Appl. Biol. 13: 109-139 Illus.

***Zweigelt, F. 1915: "Beitrage zur Kauntnis des Saugphanomens der Blattlause und der Reaktionen der Planzenzellen."

a selective influence on nutrient substances from the cell into the intercellular spaces which contain chiefly gases and water. Davidson believes that inter-cellular passage of the stylets is most generally associated with intra-cellular sucking out of the plant juices.

Dufrenoy* (1925) figures the course of the stylets of Dilachnus pinihabitans in alpine pine in connection with a discussion of the role played by this insect in the etiology of bacterious tumors. This author describes the "scar" of the aphid puncture as transversing the cells of the cortical parenchyma in a radial direction, turning in the region of the phloem to take a tangential direction. The bacterial colonies are found in the cells in the region of the tip of the puncture.

The area from which the food is extracted of course varies with the host. For example, in herbaceous plants the vascular bundles are sought after, whereas in woody stems the phloem tissues as a whole are invaded. In a coniferous stem the sap contained in the cells of the cortical parenchyma undoubtedly has some food value, but it is in the region of the phloem (sieve tubes) that there exists a naturally richer concentration of nutritive substances.

Discussion of Slide Material Examined

Altogether approximately 180 slides were mounted and examined. A great majority of this number consisted of sections from lots P-5 and P-7, since the greatest chance of finding traces of stylet tracts existed therein. No traces of these tracts were found in any of the material sectioned which was known to have been heavily infested during the 1934 season. Two explanations may be offered for this fact: One is that the sheaths left after the withdrawal of the insect proboscis are of a fragile nature and would undoubtedly be crushed in the bark where the older cells are continually being pushed outward and themselves crushed. Secondly, the chances of cuttingparallel to a sheath, even where there has been a heavy feeding, is indeed small. This is of course true for all the material. Unless the plane of cutting and the plane of penetration of stylets is exactly the same, which rests wholly on a basis of chance, there is a very small possibility of obtaining a significant length of stylet or sheath in any one section. Additional difficulty is encountered in that the sheaths frequently fall out of the tissues in the course of staining and washing, and the path so left is not clearly defined, unless one has had considerable experience in examining such material.

In approximately 20 slides out of 140 containing sections cut on the rotary microtome traces of stylets or sheaths of significant interest were found.

Dufrenoy, J. 1924: "Les Tumeurs des Resineux." Paris

Plate I is a micro-photograph of a section of tissue from lot P-5 which was fed upon by aphids during the 1935 season. Unfortunately, the outer bark tends to slough off during sectioning, thus obscuring the detail of entrance of the stylets with the host tissue. However, the stylets are clearly shown penetrating the cortex to a considerable depth. The cells in the cortex are crushed by the outward pressure of growth from the cambial region so that individual cells do not stand out as in the phloem tissues. Several cells, however, are clearly pierced by the stylets, indicating that the insect proboscis has followed an intra-cellular course in the host tissues. This indication was substantiated in all cases where individual cells stood out clearly, both in the cortex and phloem.

Plate II, after Dufrenoy (1925) is a similar view except the course of the stylets is visible to the region of the phloem where it turns to take a tangential direction.

Plate I - Stylets of Cinara schwarzii penetrating cortex of ponderosa pine.

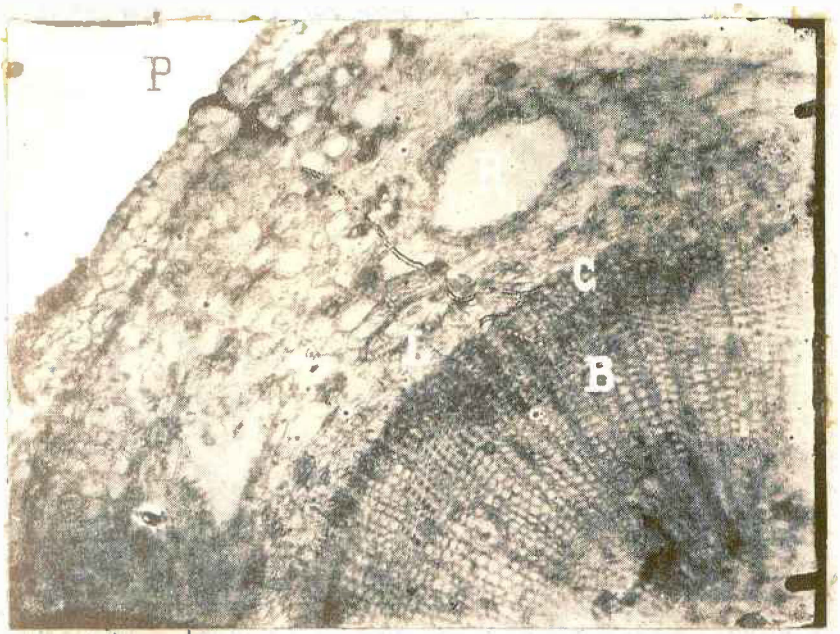

Plate II - Stylets of Dilachnus pini-habitans penetrating to cambium in Pinus halepensis (after Dufrenoy)

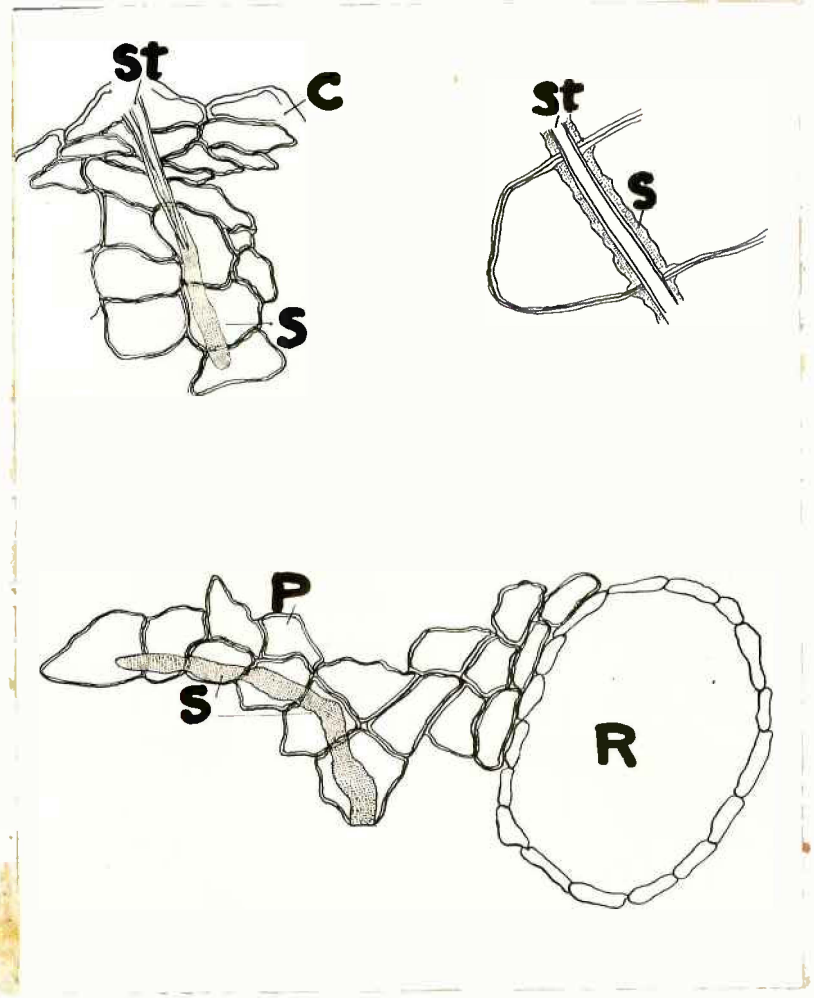

Figure 2 - Diagrammatic aspects of Stylet penetration. Top left: Course of stylet thru outer bark. Top right: Passage of stylet thru individual cell. Lower: Sheath in neighborhood of ~~cambium passing thru~~ phloem cells.

In Figure 2, diagrammatic representations are shown of some of the details of stylet penetration as seen under high power of the microscope. Top left shows the stylets entering the bark. It is difficult to say whether the cortex cells were ruptured by the insect or in course of preparation of material. The presence of the sheath, which takes a heavier stain than the surrounding cell contents, indicates that the beak penetrated the tissues and then has been partly withdrawn. Top right shows the passage of stylets directly across a cell. In no case was any rupturing or tearing of cell walls due to the passage of stylets observed. On this point indications are in agreement with Davidson and others who state that the salivary secretion dissolves a passage through the cell walls. As shown in the diagram, the sheath does not appear to go through the cell wall, although this detail is not entirely clear.

The lower diagram of Figure 2 was drawn from a portion of tissue near the phloem. In this instance the stylets have been withdrawn, leaving the sheath as an indication of their former activity. In this section the cambium runs parallel to the long axis but a few cells removed from the sheath. The turning direction indicates that the nutrient containing phloem tissues are sought after. The resin duct is included in this diagram for an expression of relative size.

In Plate III a section of sheath entering the phloem tissue cells is shown in the lower center of the photograph, with a smaller, less visible section passing through the cortex just left of the pointer on the top side. This photograph is typical of many sections obtained.

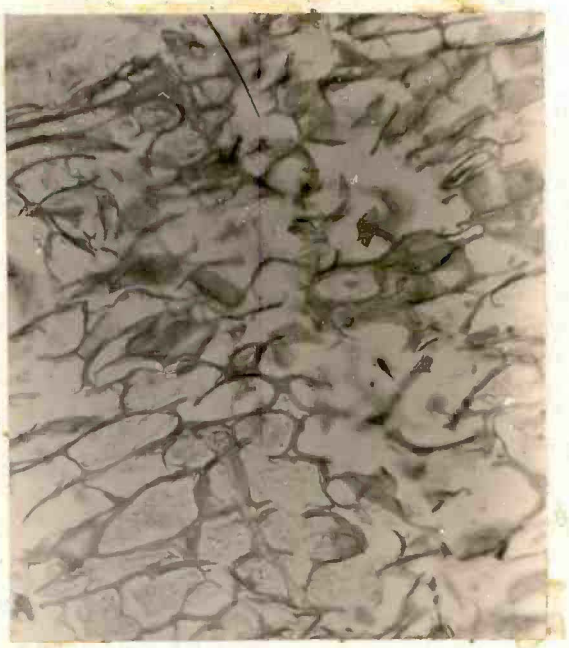

Plate III - Sheath passing thru phloem cells

Plate IV, taken with an oil immersion lens shows the tip of a sheath ending in a phloem tissue cell (center of photograph), with the cell contents taking a deeper stain than in adjacent cells. Extending toward the base, the sheath can be seen passing thru several cells. In general, no specific effects on the cells passed through by stylets could be determined.

Plate IV - Tip of sheath entering phloem cell.

Sufficient evidence was given by the slides examined to show:

1. The stylets pass intra-cellularly through the cortex to the phloem tissues.
2. The amount of mechanical damage caused is negligible.
3. A sheath is laid down wherever the stylets are moved, probably through a reaction of plant and salivary juices.
4. No irritating effect of the salivary juices on the plant tissues is evident.
5. The phloem tissues are apparently the goal of stylet penetration.

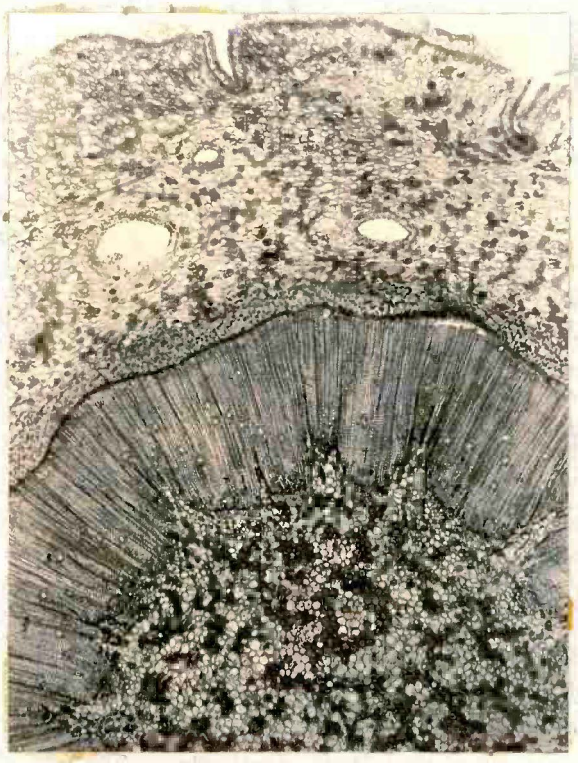

Plate V - Uninfested twig of 1935 origin.

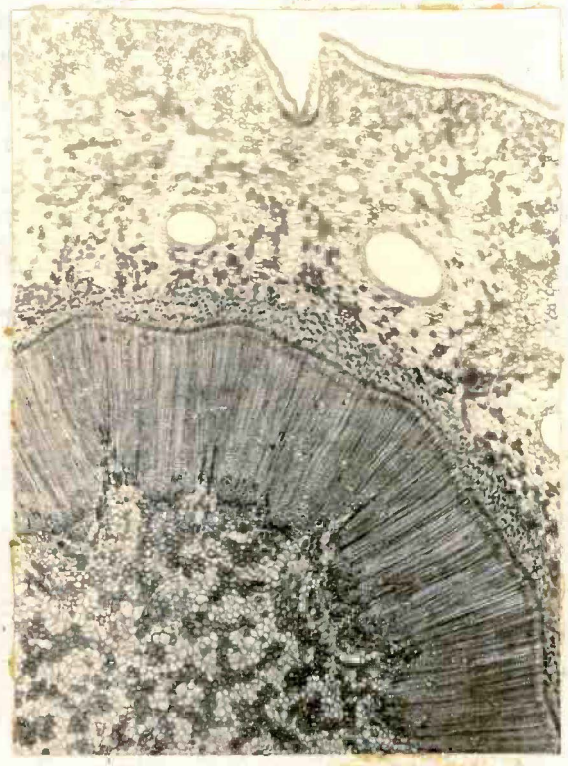

Plate VI - Infested twig of 1935 origin.

Plates V - XIV were taken with a camera objective, with sufficient magnification to give a picture of the wood structure as a whole, rather than on individual cells as obtained in the preceding plates by the use of higher magnification. Primary interst lies in determining whether or not an aphis feeding results in an injury to the wood which could be interpreted as having a detrimental effect on the tree.

Plate V is a section of a twig of 1935 origin and uninfested by aphids, and Plate VI a similar section from a twig which supported an aphid population during part of the 1935 season. Close examination of such sections was made, yet no discrimination could be made between the two.

The next series of five plates show sections from successive internodes, beginning with the 1927 growth, of a single branch known to have been heavily infested with aphis during the 1934 season but uninfested in 1935. The outstanding feature in Plate VII is a false growth ring in the 1934 growth, particularly visible in the right hand side of the photograph. It will be noted that such a false ring is also present in the 1931 growth. Such a development is brought about by a slowing down of growth and the production of thick-walled summer wood cells with subsequent proliferation of more thin-walled spring wood cells before the summer wood terminating the year's growth is laid down. It is of course impossible to say in the light of the scope of the present investigations, whether there is any connection between the aphis infestation and the production of this growth

ring. It would be necessary to examine sections from a large number of trees both infested and uninfested to determine if any connection existed. Other factors - climatic, edaphic, or physiographic might well be the cause of such a phenomenon. Since summer wood is primarily strengthening in function as opposed to the conductive function of spring wood the occurrence of a false growth ring would mean so much less tissue for conductive purposes during the season. Since under the conditions producing such an effect the tree is in a less active state this condition should probably be interpreted as an indication of some unfavorable environmental factor rather than the constitution of a physiological hindrance to the normal function of the tree.

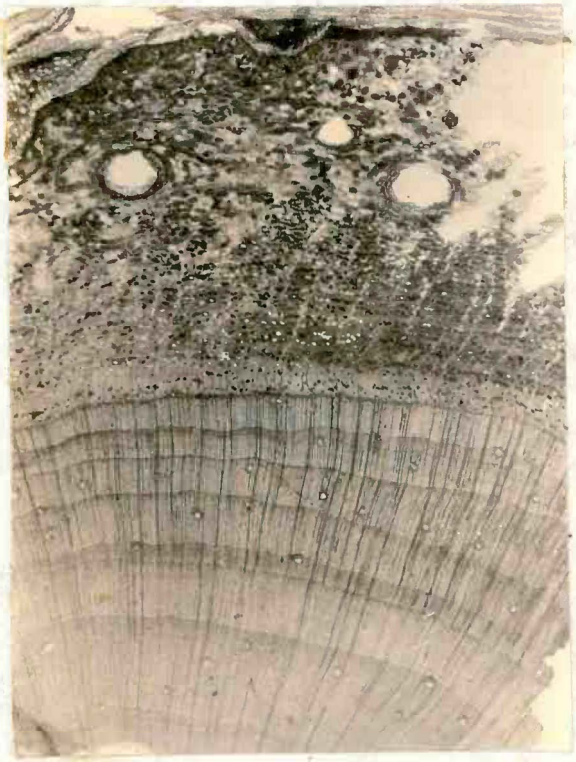

Plate VII - Infested in 1934; 1927 origin.

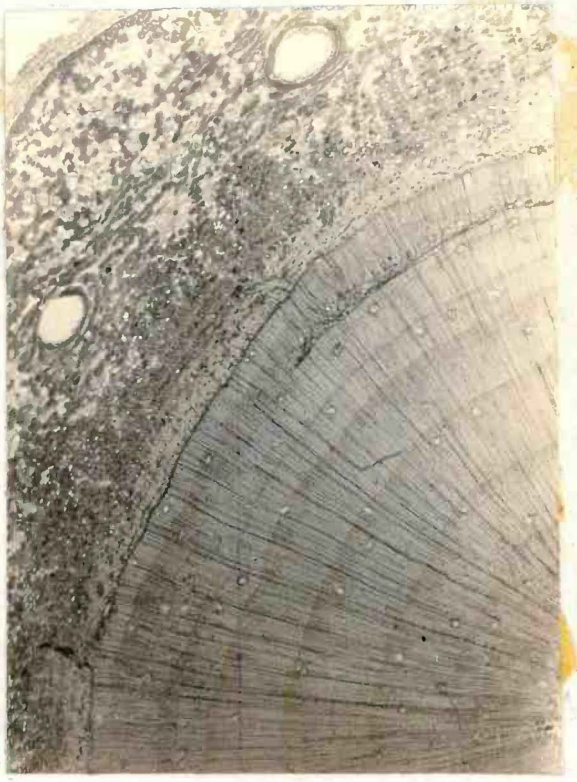

Plate VIII - Infested in 1934; 1928 origin.

In the upper right portion of Plate VIII a disturbance of the normal cell arrangement in the 1934 growth is evident. This disturbance is a total breakdown of normal structure with large inter-cellular spaces occuring, many of which are occluded by resinous or other deposits, and the crushing of many thin-walled spring wood cells. Here again it is difficult to say whether there is any connection between this injury and the aphis infestation. The false growth ring produced in 1931 is also visible here.

In Plate IX the injury in the 1934 growth, a partial 1934 false growth ring, and the 1931 false growth ring are again shown, indicating that the causal factor of these abnormalities was not completely localized in effect, as regards the longitudinal axis of the branch. As is evident from these

cross-sectional views, however, the injury does not extend completely around the stem, nor is the false growth ring consistently apparent, particularly in the 1934 growth. Such a condition would be expected if this is the result of an insect feeding. Unfortunately, there is no information as to the abundance of the aphids during the 1931 season.

Plate IX - Infested 1934; 1929 origin.

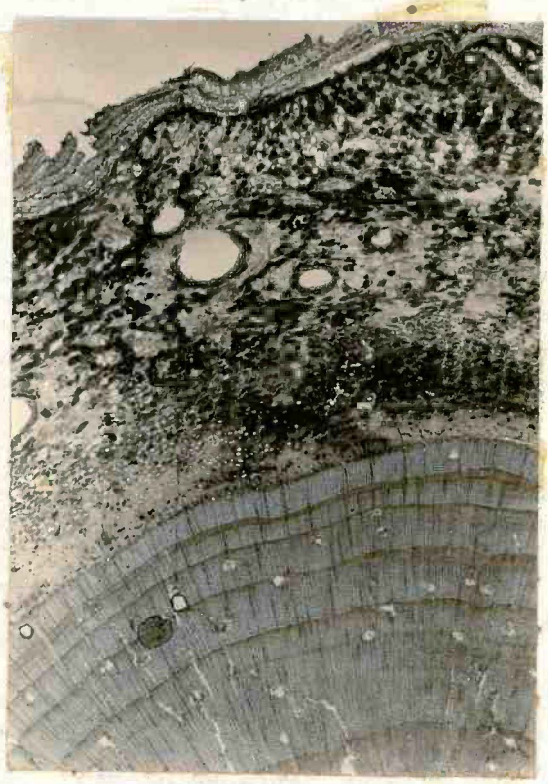

Plate X - Infested in 1934; 1930 origin.

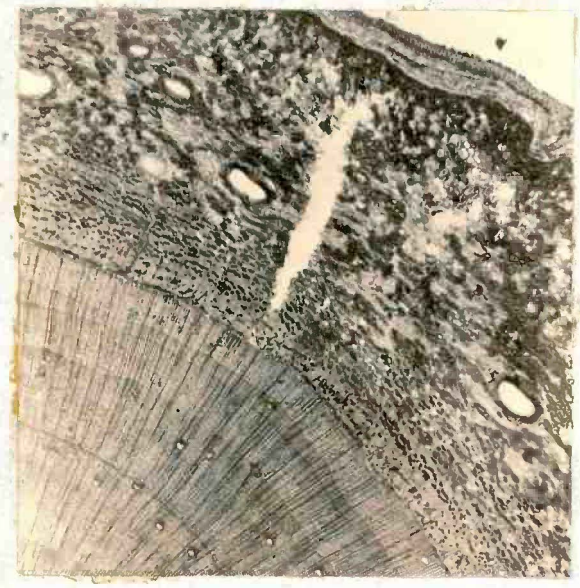

Plate X again shows the false growth rings for 1931 and 1934. In Plate XI the ring appears for 1934 but is not evident in 1931, the year of origin of this particular node. This condition may be the result of the fact that the growing tip received the most nutriment and hence is better able to withstand adverse conditions.

Plate XI (left) Infested in 1934; 1931 origin.

Plates XII and XIII show sections from adjacent internodes of another branch secured from another tree. In Plate XII a band of abnormal tissue is present in the 1931 ring and the 1934 growth is very small. In neither case, however, is a false growth ring evident. In Plate XIII the same condition exists in the 1931 growth with an area of abnormal proliferation which seems to include part of the 1930 growth. In the 1934 growth a false ring is present.

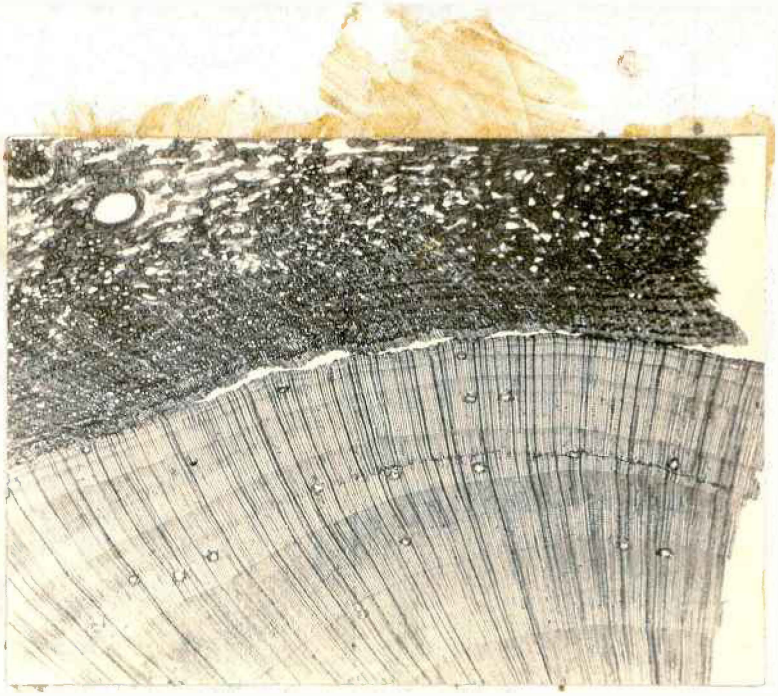

Plate XII - Infested 1934; 1925 origin.

From the limited scope of this study it is impossible to attribute any of the abnormalities depicted in the light of the result of an aphid infestation. The series of sections examined, however, has been sufficient to indicate that no serious alteration of tissue in structure, and hence indirectly in function, is exhibited as a result of aphis infestation. All indications, accordingly, are that such attack weakens the vitality of the tree according to the intensity and duration of infestation.

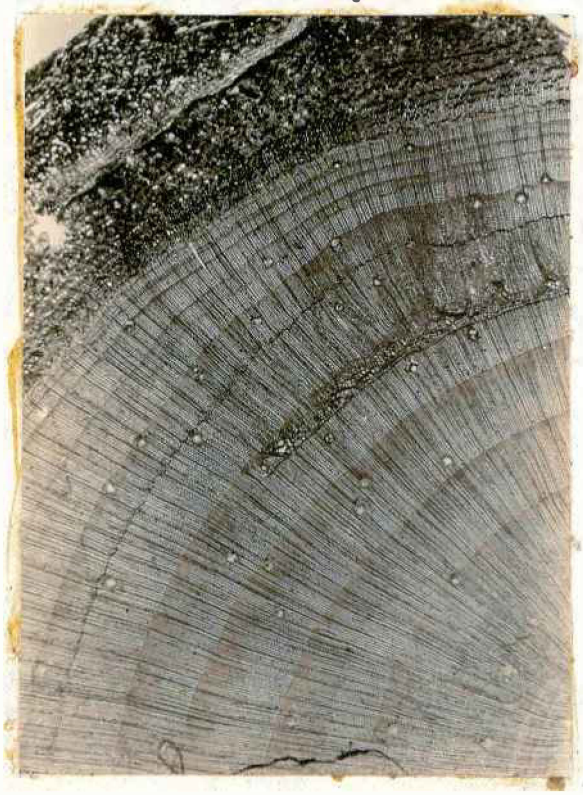

Plate XIII - Infested 1934; 1926 origin.

Since this study was undertaken in part to determine if an relation between aphis and blite exists, some sections of blited twigs were also examined. The photograph in Plate XIV is typical of sections of this material. The darkly stained elliptical areas are heavily resined, the cell lumens being completely occluded. This resining is easily observed in the field by merely slicing off the outer bark. Such an occlusion naturally interferes with the normal functions of the cells and a complete ringing of a twig by the blite infection results in flagging of the terminal portions of branches infested.

There is an ommission in the series of plates in that no pictures of normal growth are shown other than Plate V which is of a twig of the current year. Sections of older twigs were cut but crushed considerably. In the material examined no injury or false growth rings were detected. It would of course be necessary to examine a large number of trees both infested and uninfested, before the abnormalities described could be definitely ascribed to aphis attack.

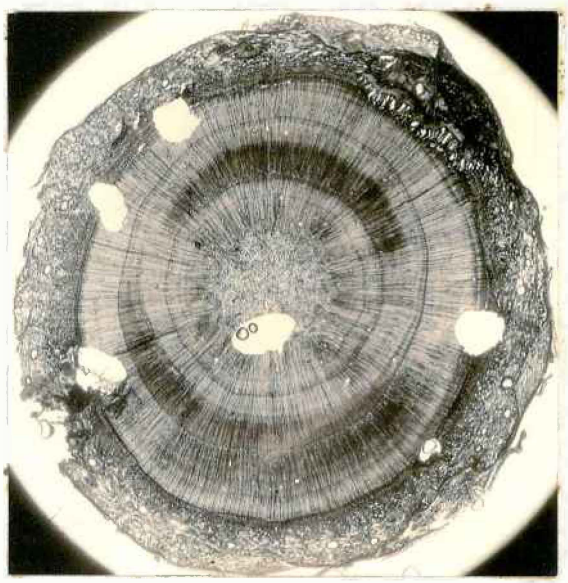

Plate XIV - Resined condition of blite infected fourth year twig.

It may be well to restate here that this entire report is of a preliminary nature. In particular does this apply to the histological investigations. The material used for this purpose was not as specifically selected as would be necessary for detailed study. The preparations made have been suffieient, however, to give evidence to the question of blite and aphid relationship. It is not suggested that they are thorough enough to enable a detailed discussion of the various tissues entered by the insect stylets and the effect of this disturbance on cellular structure and function. Hence no conclusions are attempted, as to physiological relationships although it is evident that a heavy aphis infestation must result in a drain on the vitality of the tree which in turn would be expressed by lessened growth. A detailed investigation into the nature of the effect of various sucking insects on forest trees is a study to be desired; such a contribution to the field of forest entomology would fill what is at the present time a regretted lacuna on the literature.

ACKNOWLEDGEMENTS

Appreciation of assistance and cooperation outside the Bureau is expressed to the following:

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SUMMARY

An investigation of the aphis situation on the Prescott National Forest was conducted during the period May 9 - August 1, 1935. Two species of needle aphis, Schizolachnus pini-radiatae and Cinara sp., a woolly aphid, Chermes sp., and a stem aphid, Cinara schwarzii was most abundant and was the form which was epidemic on the forest in 1934. It was definitely established that the stem aphid was nowhere present in as great abundance as during the previous year. The other aphis species were exceedingly sporadic in appearance and distribution and were of no particular concern.

Cinara schwarzii was first found on June 17 and reached a peak of abundance during the first week of July. Infestations were found in three localities on the forest. In contrast to the 1934 epidemic when the infestation was widespread and trees of all sizes were infested, the infestation during the current year was never at any time heavy, and was confined entirely to young trees in open areas. With few exceptions the infestations were limited on the tree to the current year's growth. In the latter part of July the species rapidly disappeared until but a few isolated colonies could be located on the first of August.

This species was successfully reared in a field cage. Early in July the aphids began moving back from the growth of the current year onto the older growth, and colonies wereestablished as far back as the 1932 growth. On this older growth the colonies were more dense and produced winged individuals in considerable nnmbers. Winged forms likewise produced young and established colonies when confined to a section of branch. Everywhere ants were in attendance on the aphid colonies, apparently living with the aphis in symbiosis for the purpose of feeding on the "honey dew".

From observations made in the course of the season's study there seems practically no possibility of any direct connection between the stem aphis and the twig blite which is prevalent in the Prescott forest. The distributional areas of infested and infected trees do not coincide, and trees which were heavily infested in 1934 are of normal appearance during the current season, eliminating the theory that aphis feeding causes an injury similar to that of the blite.

A histological study has been made of aphis infested twigs and comparison made with normal and blited twigs. From these observations it is evident that the aphid feeding punctures do little if any damage to the wood tissues. The stylets penetrate into the phloem region with no evidence of an irritating effect of the salivary fluid which probably dissolves the intra-cellular path for the stylets. A sheath laid down surrounding the stylets demonstrated other than the occurrence of a false growth ring there is no abnormality which can be possibly attributed to the insect attack.

Comparison with sections of blited twigs bear out the conclusions of field observations, namely; there is to date no basis on which a relation between blite and aphis infestations can be established. The typical resining in blited twigs has no counterpart in aphis infested twigs, nor were any feeding punctures found in blited twigs.

A more detailed histological investigation would enable the determination of the specific effect of feeding punctures and possibly some of the physiological reactions involved.